

Different Activities of Antibacterial Waste from Agung

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Different Activities of Antibacterial Waste from Agung Semeru Varieties Lumajang towards *Staphylococcus aureus* and *Pseudomonas aeruginosa*

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ABSTRACT

Banana peel of Agung Semeru, varieties Lumajang contains bioactive compounds that function as antimicrobial, namely Phenol, Terpenoid / steroid, saponin and Alkaloid. The purpose of this research is to know the difference of banana extract of Agung Semeru varieties of lumajang, in inhibiting the growth of *Staphylococcus aureus* and *Pseudomonas aeruginosa*, and to know the optimal concentration which can inhibit the growth of them. The method used was four of factors, that is: 0, 25, 50, 75 and 100 %. Each treatment was repeated three times. The results show that there is an effect of giving extract of Semeru banana peel ($\text{sig} = 0,00$) to *Staphylococcus aureus* and *Pseudomonas aeruginosa* growth at various concentration (0%, 25%, 50%, 75% and 100%). For extract of Semeru banana peel at concentration 75% ($3,19 \pm 0,84$ mm) showed better result from other concentration to inhibiting the grow of *Staphylococcus aureus* and *Pseudomonas aeruginosa*.

Keywords: Agung Semeru Lumajang, *Staphylococcus aureus*, *Pseudomonas aeruginosa*

INTRODUCTION

Banana peel is one of the natural resources that have potential to be herbal medicine. This is in accordance with the research result that has been done by Indrawati, et al. (2015) that the water extract of Ambon's Banana peel can lower blood sugar levels because there are bio-active compounds such as flavonoid, fenolik, sapones, based on the other research results about Banana peel i.e. Ambon banana also contain bio-active compounds that is fitochemical component which potentially functioned as anti-microbe such as flavonoid 5,6,7,4'-tetrahydroxy-3,4-flavon-diol (Atun, et al., 2007), phylobatatin, antrakuinon and kuinon (Salau, et al., 2010). Banana peel extract also have the compound of secondary metabolites results that potentially becomes anti-fungus compounds i.e. fenolik and terpen category (Viyekanand, 2011), alkaloid and triterpenoid (Pratiwi, 2011).

Agung Semeru Banana, Lumajang variety is one of the banana species which becomes the characteristics of Lumajang districts, where the demand of Agung Semeru banana is growing in each year. Based on the production data in 2008, it showed that Agung Semeru Banana production was 50.776,2 tones and keeps growing its production (Fiqrotul, 2011). Based on the research result that has been done by Sari and Susilo (2017), shows that extract banana peel of Agung Semeru Lumajang variety contain various bio-active

compounds which have functions as anti-microbe compound, that is fenol compound, terpenoid/steroid compound, saponin and alkaloid compound.

On the other hand, the increasing amount of Agung Semeru banana production in Lumajang district which is mostly harnessed its fruit to be Agung banana chips, causes the stockpiling of trash which are mostly in the form of Banana peel waste. Agung Semeru Banana peel waste are still considered as organic waste and do not contaminate the environment, and harnessed as animal feed such as goat, cow, or even buffalo (Susanti, 2006). Thus, it is necessary to conduct a more economical usage, concerning that this banana peel of Agung Semeru Banana Lumajang variety have antimicrobe compounds so that it can be used as natural medicine to suppress the growth of pathogen microbes which causes infection diseases as well as to be one of the solutions which is used to decrease the resistance of pathogen micro-organism towards synthetics medicines.

Infection diseases are one of the diseases that mostly suffered by Indonesian people which caused by micro-organism. One of the micro-organism that causes infection diseases that is from bacterial group such as *Pseudomonas aeruginosa* (Negative Gram Bacterial) and *Staphylococcus aureus* (Positive Gram Bacterial) (Djide and Sartini,

2008). The increasing of micro-organism resistance even bacteria or fungus towards artificial antibiotics, so another alternative is needed to decrease that resistance, one of the ways is by utilizing natural compounds that are available in the plant in the form of secondary metabolites which is called by phytochemical compounds. The purpose of this research is to know the potential of Agung Semeru Banana peel extract towards Positive Gram bacteria (*Staphylococcus aureus*) and Negative Gram Bacteria (*Pseudomonas aeruginosa*), as well as to know the optimum concentrate that can inhibit the growth of *Staphylococcus aureus* and *Pseudomonas aeruginosa*.

MATERIALS AND METHODS

Banana peel Extraction

It is started by cleaning Agung Semeru Banana peel until there is no dirt that sticks to it and cutting it into tiny little pieces. After that, it is dried by wind until the Banana peel is dry. Next, the dried Agung Banana peel should be inserted into the blender and blend it until it becomes smooth and then it is macerated. At approximately 100 gram of Agung Semeru Banana peel that has been smoothed is macerated with 300 ml of water for 1 x 24 hours. The extract that is acquired is then filtered by using Buchner funnel and using vacuum and filtrate that is obtained and it is steamed by rotary vacuum evaporator so that condensed extract is acquired. To get the Banana peel extract in large amount, so that the extraction process is conducted for six times (Supriyanti et al., 2015).

The Test of Extraction Activity

The test of Agung Semeru Banana peel ethanol anti-bacterial power is tested by using disc paper diffusion method. The suspension of tested bacteria is 0.5 ml and it is poured in petri dish then pours NA media 10 ml in sterilized condition, make it homogeny, and let it be solid. Agung Semeru Banana peel extract that has been diluted using distilled water with 100, 50, 25, 12.5, 6.2, 3.1 mg/mL concentrate, and then each extract concentrate is inserted in reaction tube. 3 empty paper discs with 14.20 mm size which has been

sterilized are inserted in each reaction tube and let it be in 10 minutes, and next by paper pin-set, paper disc are drained on the edge of the tube in 10 minutes. Put the paper disc that has been drained by using pin-set. As control, it is necessary to put paper disc that have been saturated in aquadest liquid, and as a comparison, antibiotics tetracycline 30 µg/disc paper disc are used. The test is done three times repetitions (triple) so that each dish has 3 paper discs. The dish that has the paper disc is then incubated in 37°C for 18 – 24 hours. The observation of anti-bacterial power test is done by measuring the diameter of resistance area (DHP) bacteria growth that is formed in around paper disc with calipers. The measurement is done in millimeter (mm). The positive test is marked by the formation of clear halo (hindrance area) in around paper disc after incubation period, this shows the existence of anti-microbes activities (Meilina, 2015 and Poeloengan et al, 2007).

Data Analysis

The data that is observed covers inhibition zone diameter of agung semeru Banana peel extract and mas kirana banana lumajang variety which is analyzed quantitatively by using SPSS 20. The result of the observation data will be analyzed by using one-way ANOVA 5% and it is continued by Duncan's multiple range test (DMRT) in order to compare between treatments. Before ANOVA test is conducted, the data is first tested by using normality and homogeneity tests.

RESULTS

This research has purpose to know the different activity of anti-bacteria from agung semeru Banana peel extract lumajang variety towards the growth of *Pseudomonas aeruginosa* and *Staphylococcus aureus* bacteria. Based on the research that has been conducted, it shows that Agung Semeru Banana peel extract Lumajang variety in various concentrates i.e. 0%, 25%, 50%, 75% and 100% shows the result that there is a significant different towards the growth of good *Staphylococcus aureus* bacteria, even *Pseudomonas aeruginosa* (Picture 1).

ANOVA						
		Sum of Squares	df	Mean Square	F	Sig.
Diameter Zona Hambat Pertumbuhan <i>Staphylococcus aureus</i>	Between Groups	801,807	4	150,402	82,084	,000
	Within Groups	24,222	10	2,422		
	Total	826,028	14			
Diameter Zona Hambat Pertumbuhan <i>Pseudomonas aeruginosa</i>	Between Groups	557,426	4	139,357	52,897	,000
	Within Groups	28,345	10	2,835		
	Total	585,771	14			

Picture 1. The statistics test by using ANOVA 5%, Agung Semeru Peel of Banana Extract towards *Staphylococcus aureus* and *Pseudomonas aeruginosa* bacteria growth zone.

Based on the result of statistics analysis by using ANOVA one-way test that has 5% level of trust show that there is a significance between treatments ($\alpha = 0.00$) (Picture 1), where the best treatment for Agung Semeru Banana extract Lumajang variety that existed in the treatment with 100% concentrate ($17,55 \pm 3,39a$) in inhibiting the growth of *Pseudomonas aeruginosa* bacteria and 100% concentrate ($19,25 \pm 0,91c$) in inhibiting the

growth of *Staphylococcus aureus* Positive Gram bacteria (Table 1). The provision of Agung Semeru Banana peel extract Lumajang variety shows that there is a significant difference between treatment of various concentrate towards the growth of *Pseudomonas aeruginosa* bacteria even *Staphylococcus aureus* bacteria and shows that the higher the concentrate given, the larger the obstacle zone diameter that is formed (Table 1; Picture 2).

Table 1. The obstacle zone diameter of Agung Semeru Peel of Banana extract Lumajang variety towards *Staphylococcus aureus* and *Pseudomonas aeruginosa*

Extract Concentrate (%)	The average diameter of obstacle zone (mm)	
	<i>Pseudomonas aeruginosa</i>	<i>Staphylococcus aureus</i>
0	$0,00 \pm 0,00^a$	$0,00 \pm 0,00^a$
25	$12,62 \pm 1,03^b$	$11,83 \pm 1,25^b$
50	$14,02 \pm 0,40^{bc}$	$13,2 \pm 1,28^b$
75	$14,62 \pm 0,68^b$	$13,8 \pm 2,84^b$
100	$17,55 \pm 3,39^a$	$19,25 \pm 0,91^c$



Picture 2. The graph shows the relationship between obstacle zone diameter (mm) of Agung Semeru Peel of Banana extract towards the growth of *Staphylococcus aureus* and *Pseudomonas aeruginosa* with the concentrate of various treatments

In table 1, for the provision of Agung Semeru Banana peel extract, it shows that the treatment with 25% ($12,62 \pm 1,03b$), 50% ($14,02 \pm 0,40bc$), 75% ($14,62 \pm 0,68b$) and 100% ($17,55 \pm 3,39a$) concentrate show better results in comparison to control ($0,00 \pm 0,00a$), with 100% concentration treatment, it shows more results that is marked by growth inhibit zone diameter of *Staphylococcus aureus* even *Pseudomonas aeruginosa* which is way bigger. The anti-bacteria activity of Agung Semeru Banana peel extract looks like all treatment concentration started from the smallest concentrate to the biggest concentrate (25%, 50%, 75% and 100%) even for *Pseudomonas aeruginosa* and *Staphylococcus aureus* bacteria with the best concentrate, it is existed in the extract treatment with 100% concentrate. While concentrate treatment which shows the average changing of inhibit zone growth diameter of *Pseudomonas aeruginosa* even *Staphylococcus aureus* bacteria towards Agung Semeru Banana peel extract can be seen in Picture 2.

Based on the research result, it shows that the provision of Agung Semeru Lumajang Banana peel extract has anti-microbes activity in inhibiting positive Gram bacteria *Staphylococcus aureus* and Negative Gram bacteria *Pseudomonas aeruginosa* in comparison to the negative control treatment. Based on the statistical analysis by using ANOVA 5%, it shows that the provision of Agung Semeru Banana peel extract towards various concentrates (25%, 50%, 75% and 100%) indicates a significant level towards obstacle zone diameter of bacteria, either *Pseudomonas aeruginosa* or *Staphylococcus aureus*. For the advanced analysis test which uses Duncan Test 5% indicates the different effect of providing Agung Semeru Banana peel extract between various treatment concentrates, where according to Simanjutak (2008) that Duncan test is used to see which treatment that has the same effect or different and even from the smallest effect to the biggest effect from one another.

Based on the anti-bacteria power resource according to Davis and Stout (1971), it is as follows: Inhibit Zone Diameter (DZP) > 5 mm is categorized as weak, DZP 5-10 mm is categorized as medium, DZP 10-20 mm is categorized as strong and DZP > 20 mm is the strongest category. Based on that criteria, so that the anti-bacterial resource of Agung Semeru Banana peel extract in this research is considered to be powerful in inhibiting Negative Gram bacteria *Pseudomonas aeruginosa* and Positive Gram bacteria *Staphylococcus aureus* that can be seen in table 1 and picture 1. The extraction ability of Agung Semeru Banana peel in inhibiting

the growth of bacteria is because despite there is fitochemical compound (Sari and Susilo, 2017) that is anti-bacteria is also because of the structure of both bacteria (Radji, 2011).

In this research, inhibit zone diameter of Agung Semeru Banana peel extract is way bigger in inhibiting the growth of Positive Gram bacteria *Staphylococcus aureus* which is compared to Negative Gram Bacteria *Pseudomonas aeruginosa*. This is caused by there is a difference of cell wall structures for both bacteria. The structure of cell wall from Positive Gram bacteria i.e. *Staphylococcus aureus* consists of peptidoglikan layer that is thicker and forms thicker structure and firm as well as teikoat acid as one of the cell wall substance. While for Negative gram bacteria cell wall, it contains peptidoglikan layer which is thin with on the outer part of peptidoglikan there is membrane as well as do not contain teikoat acid so that it is reluctant to the physical environment condition and chemical materials.

According to Schlegel (1994), the ability of anti-microbes material in killing or inhibiting the growth of certain micro-organism depends on the compounds concentrate/microbes materials itself. In this research, based on the advanced test i.e. Duncan and curv in Picture 1, shows that zone diameter inhibit the growth of *Staphylococcus aureus*, and *Pseudomonas aeruginosa* bacteria with extract 100% concentrate bigger than extract concentrate 25%, 50% and 75%. This shows that the bigger the extract concentrate of Agung Semeru Banana peel which is given, the bigger inhibit zone diameter that are formed, this is because concentrate factor, anti-microbes materials which really influence and also decide the ability in inhibiting the growth of bacteria (Ajizah, 2004).

CONCLUSION

Agung Semeru Lumajang variety Banana peel extract has an activity as anti-microbes so that the provision of Agung Semeru Banana peel extract (sig = 0,00) in various concentrate (0%, 25%, 50%, 75% and 100%) shows the significantly affected result towards the growth of Negative Gram material *Pseudomonas aeruginosa* and Positive Gram bacteria *Staphylococcus aureus* with 100% concentrate is the best concentrate in inhibiting the growth of *Pseudomonas aeruginosa* bacteria ($17,55 \pm 3,39a$) and Positive Gra *Staphylococcus aureus* bacteria ($19,25 \pm 0,91c$).

CONFLICT OF INTEREST

This research was carried out without any element of conflict of interest.

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AUTHOR CONTRIBUTION

DNRS contributes as a research design maker and is responsible for the results of the research, the DKS assists the research, and SDA analyzes the research data. the entire research team has read and approved the final contents of the article.

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